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January 8, 2004

BEHQ-80-373 AR 226-1552

Document Processing Center EPA East – Room 6428 Attn: Section 8(e) Office of Pollution Prevention and Toxics US EPA

1200 Pennsylvania Avenue NW Washington DC 20460-0001

RE: TSCA 8(E) SUPPLEMENTAL SUBMISSION:

Docket No. 8EHQ-0602-15091

Dear Docket Coordinators:

3M has previously informed the EPA (February 28, 2002 and April 3, 2002) of the results of an oral gavage two-generation reproduction study in rats with ammonium perfluorooctanoate (CAS# 3825-26-1). In today's submission, 3M provides results from subsequent related testing with this material. This information has been previously submitted to and discussed with EPA.

Enclosed please find the final study titled "Ammonium Perfluorooctanoate: Age Effect on the Plasma Concentration in Post-Weaning Rats Following Oral Gavage."

Please contact John Butenhoff (651-733-1962) if you have any questions or if we can provide additional information.

Sincerely,

Katherine E. Reed

Staff Vice President, Environmental, Health and Safety Operations

Enclosure

c:

Ms. Andrea Malinowski DuPont Building 1007 Market Street Wilmington, DE 19898

atherine E. Keed

Dr. Robert Rickard DuPont Haskell Laboratory Elkton Road, P.O. Box 50 Newark, DE 19714-0050 ⊕ EPA-OTS

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AR226-1553

Study Title

Ammonium Perfluorooctanoate: Age Effect on the Plasma Concentration in Post-Weaning Rats Following Oral Gavage

AUTHOR: Xing Han, Ph.D.

STUDY COMPLETED ON: December 15, 2003

PERFORMING LABORATORY: E.I. du Pont de Nemours and Company

Haskell Laboratory for Health and Environmental Sciences

Elkton Road, P.O. Box 50 Newark, Delaware 19714-0050

LABORATORY PROJECT ID: DuPont-13267 T-6889.10

WORK REQUEST NUMBER: 14762

SERVICE CODE NUMBER: 1389

SPONSOR: E.I. du Pont de Nemours and Company

Wilmington, Delaware 19898

U.S.A.

and

3M Company 3M Center Building St. Paul, MN 55144-1000

GOOD LABORATORY PRACTICE COMPLIANCE STATEMENT

This study was conducted in compliance with U.S. EPA TSCA (40 CFR part 792) Good Laboratory Practice Standards.

Study Director:

Research Toxicologist
E.I. du Pont de Nemours and Company

QUALITY ASSURANCE STATEMENT

Haskell Sample Number(s):

24921

Dates of Inspections:

Protocol: June 18, 2003

Conduct: July 8,9,21,28,30, 2003

Records, Reports: October 27-30, 2003

Dates Findings Reported to:

Study Director: June 18, 2003; November 3,11, 2003

Management: November 3,11, 2003

Reported by: (1) on da) K. Kelly Wonda W. Kelly

Quality Assurance Auditor

- 3 -

CERTIFICATION

We, the undersigned, declare that this report provides an accurate evaluation of data obtained from this study.

Research Manager and Director

Issued by Study Director:

Xing Han, Ph.D.

Research Toxicologist

15-Dec-2003

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STUDY INFORMATION

9th Collective Nomenclature: Octanoic acid, pentadecafluoro-, ammonium salt

Synonyms/Codes: • Am

• Ammonium perfluorooctanoate

FC-143 FLUORAD Brand Fluorochemical Surfactant

(3M Company, Specialty Materials)

• C-8

• Perfluorooctanoate, ammonium salt

PFOA

• H-24921

• Lot 332 (3M Specialty Materials) (Lot No.)

Haskell Number: 24921

CAS Registry Number: 3825-26-1

Purity: 95.2% – 97.99%

Straight chain: 77.6%

Branched: 12.6% internal monomethyl (non-alpha)

9% isopropyl
0.2 % tert-butyl
0.1% gem-dimethyl
0.1% alpha monomethyl

Known Impurities: C₄ (C₃F₇CO₂ NH₄), 0.01%

C₅ (C₄F₉CO₂ NH₄⁺), 0.03% C₆ (C₅F₁₁CO₂ NH₄⁺), 0.43% C₇ (C₆F₁₃CO₂ NH₄⁺), 0.57% C₉ (C₈F₁₇CO₂ NH₄⁺), 0.16% Monohydro APFO, 0.09% Monounsaturated APFO, 0.72%

Undefined (possibly) substituted perfluorocyclo species,

0.2% cyclopentyl, and 0.1% cyclohexl

Physical Characteristics: White solid

Stability: The test substance appeared to be stable under the

conditions of the study; no evidence of instability was

observed.

Sponsor: E.I. du Pont de Nemours and Company

Wilmington, Delaware 19898

U.S.A.

and

3M Company 3M Center Building St. Paul, MN 55144-1000

Experimental Start/Completion: June 30, 2003 / August 7, 2003

Study Initiated/Completed: June 27, 2003 / (see report cover page)

STUDY PERSONNEL

Study Director: Xing Han, Ph.D.

Primary Technicians: LaRue A. Manning, B.A.

Brian P. Shertz, A.A.

Management: Matthew S. Bogdanffy, Ph.D., D.A.B.T.

Gary W. Jepson, Ph.D.

Toxicology Report Preparation: Lisa G. Burchfield, A.A.

Management: Nancy S. Selzer, M.S.

Laboratory Veterinarian: Thomas W. Mayer, D.V.M., Diplomate A.C.L.A.M.

Management: Janice L. Connell, M.S., B.A., C.I.H.

SUMMARY

The relationship between age and plasma concentration of ammonium perfluorooctanoate (PFOA) in rats has been investigated in this study. Male and female rats between 4 and 8 weeks old were administered 10 mg/kg of PFOA by single oral gavage. The plasma concentration of PFOA at 24 hours post-dose was measured by LC/MS. The results showed that the plasma concentration of PFOA in rats was dependent on the age and sex of the rats.

A. Age Effect

In both male and female rats, the greatest age-related difference in PFOA plasma concentration was observed between 4 and 5 weeks of age. The 5-week old male rats had PFOA plasma concentrations that were 5.4-fold higher than the 4-week old male rats. In contrast, the PFOA plasma concentrations in the 5-week old female rats were 2.4-fold lower than in the 4-week old rats. PFOA levels in plasma were no longer significantly affected by age in rats of either sex that were older than 5 weeks (through study termination at 8 weeks).

B. Sex Comparison

At 4 weeks of age, male rats had PFOA plasma concentrations that were 2.7-fold higher than in females. The PFOA plasma concentrations in 5- to 8-week old male rats were 34.7- to 65.1-fold higher than that in the females of the same age.

INTRODUCTION

Ammonium perfluorooctanoate (PFOA) is a perfluorinated octanoic acid salt used as an industrial surfactant. Elimination of PFOA in rats is sex-dependent and hormone regulated. PFOA elimination is much faster in female rats than in male rats, (1) down-regulated by testosterone in both female and castrated male rats, (2,3) and up-regulated by estradiol in the male rats. However, these studies were conducted on sexually mature rats. In the current study, male and female post-weaning rats at different ages were dosed with PFOA, and the plasma concentrations of PFOA at 24 hours post-dosing were compared. This post-dosing time point was chosen because 24 hours after dosing, the sex difference in PFOA elimination in mature rats can be easily distinguished via the plasma concentration of PFOA. (5) The objective of this study was to determine if immature male and female rats eliminate PFOA from blood at rates different than those observed in mature rats. This study was not designed to fully characterize the pharmacokinetics of PFOA in immature rats, and was not designed for use in determining definitive elimination rate constants.

MATERIALS AND METHODS

A. Test Substance

PFOA (ammonium perfluorooctanoate, FC-143, Lot 332) was obtained from 3M (St. Paul, MN) and assigned Haskell Laboratory Number H-24921 upon receipt. Available information on the purity, composition, contaminants, synonyms, hazards, and hazardous material classification(s) was provided by the vendor and documented in the study records and report.

CAS Registry Number:

335-67-1

Molecular Weight:

414.1

Molecular Formula:

 $C_8F_{15}O_2H$

B. Test System

Male and female Crl:CD[®](SD)IGSBR rats were obtained from Charles River Laboratories, Inc., Raleigh, North Carolina. The Sprague-Dawley rat was chosen for this study because of the extensive experience with this strain and its suitability with respect to longevity, sensitivity, and low incidence of spontaneous diseases. Furthermore, the Sprague-Dawley rat has been used previously for toxicokinetic testing of PFOA and other fluorinated test materials.^(2,5,6)

At the time of dosing, rats were 4, 5, 6, 7, and 8 weeks of age and the weight variation did not exceed $\pm 20\%$ of the mean weight by dose group and sex. Information on the cage labels

included the Haskell animal number and an individual identification number assigned to each rat. The individual identification number was placed on the tail of each rat.

C. Animal Husbandry

1. Animal Housing

Upon arrival at Haskell Laboratory, rats were removed from shipping cartons and housed in appropriate cages according to Standard Operating Procedures. Animals were maintained under quarantine for at least three days, had at least one recorded weight gain, and no abnormalities detected. After the quarantine period, rats were selected for study.

Throughout the dosing period with test substance, the rats were housed individually in appropriate cages according to the SOP.

2. Environmental Conditions

Animal room(s) were maintained at a temperature of 18-26°C (targeted to 22-24°C) and a relative humidity of 30-70% (targeted to 40-60%). Animal rooms were artificially illuminated (fluorescent light) on an approximate 12 hour light/dark cycle. Unless judged by the study director or the laboratory veterinarian to have significantly affected the results of the study, the relative humidity and temperature ranges in the housing rooms was recorded but not included in the final report.

3. Feed and Water

All animals were provided tap water ad libitum and fed PMI[®] Nutrition International, LLC Certified Rodent LabDiet[®] 5002 ad libitum. Animals were fasted overnight for approximately 12 hours before dosing with test substance. Food was returned approximately two hours post-dose.

4. Animal Health and Environmental Monitoring Program

As specified in the Haskell Laboratory animal health and environmental monitoring program, the following procedures are performed periodically to ensure that contaminant levels are below those that would be expected to impact the scientific integrity of the study:

- Water samples are analyzed for total bacterial counts, and the presence of coliforms, lead, and other contaminants.
- Feed samples are analyzed for total bacterial, spore, and fungal counts.
- Samples from freshly washed cages and cage racks are analyzed to ensure adequate sanitation by the cagewashers.

Certified animal feed is used, guaranteed by the manufacturer to meet specified nutritional requirements and not to exceed stated maximum concentrations of key contaminants, including

specified heavy metals, aflatoxin, chlorinated hydrocarbons, and organophosphates. The presence of these contaminants below the maximum concentration stated by the manufacturer would not be expected to impact the integrity of the study.

The animal health and environmental monitoring program is administered by the attending laboratory animal veterinarian. Evaluation of these data did not indicate any conditions that affected the validity of the study.

D. In-Life Methods

1. Animals

The experimental groups were as shown below:

| | Ag | e* | Numbe | r of Rats |
|-------|-------|------|-------|-----------|
| Group | Weeks | Days | Male | Female |
| I | 4 | 28 | 10 | 10 |
| II | 5 | 35 | 10 | 10 |
| III | 6 | 42 | 10 | 10 |
| IV | 7 | 49 | 10 | 10 |
| V | 8 | 56 | 10 | 10 |

^{*} Animals were ±1 day from the target age at the time of dosing. The actual age at dosing is documented in the study records.

2. Dose Preparation, Analysis, and Rates

The test substance was administered to each rat as a single oral gavage. This route was chosen because it is the route most commonly used for toxicity studies of PFOA. PFOA was mixed with HPLC grade water to achieve the target dose concentrations. For all experiments, a single dose level was used equivalent to 10 mg PFOA/kg body weight, and the dose volume was approximately 4 mL/kg body weight. Dose solutions were prepared one day before the day of use and stored in a 4°C refrigerator prior to use.

LC/MS methods that were used for dose verification are described in Section E, Analytical Methods.

3. Sacrifice and Blood Sampling

Rats were sacrificed by CO₂ asphyxiation and exsanguinated by cardiac puncture.

4. Experimental Procedure

Rats were administered PFOA as a single oral dose of 10 mg/kg. Twenty-four hours after PFOA administration, rats were sacrificed and whole blood samples were collected by cardiac puncture. Blood samples were all collected within 20 minutes of the desired sample time. Whole blood samples were placed into EDTA tubes and maintained on wet ice. Plasma was separated by centrifugation and frozen at <-10°C until analysis.

Kidneys and livers were collected at sacrifice and were flash frozen in liquid nitrogen and stored at <-60°C for possible future biochemical analysis.

E. Analytical Methods

1. Verification of Dose Solution

PFOA standards (1.0, 0.5, 0.2, 0.1, and 0.05 μ g/mL) were prepared in HPLC-grade water. The dose solution was diluted by 10,000-fold prior to the analysis by LC/MS method.

2. Extraction of PFOA from Plasma for LC/MS Analysis

Plasma samples were processed by protein precipitation (PPT) using Isolute Array protein precipitation columns (Jones Chromatography, Lakewood, CO). A 0.5 μ g/mL solution of perfluorononanoic acid (Aldrich Chemicals, Milwaukee, WI) in acetonitrile (ACN) was used as an internal standard for quantitation of PFOA. Plasma samples were thawed, and a 20 μ L aliquot of each sample was applied to the PPT array. The plasma samples were precipitated by adding appropriate dilution rate volumes of ACN/internal standard solution to the PPT array. Dilution rates, ranging from 1:4 (60 μ L of internal standard solution) to 1:50 (980 μ L of internal standard solution), were utilized in order to capture the sample concentrations within the standard curve parameters. The array was slowly eluted under vacuum into a 96-well receiver plate, centrifuged at ~3000 rpm for 5 minutes, and extracts were analyzed by LC/MS.

3. Instrumentation

| System: | - | hromatograph, equipped with quaternary pump, column |
|--------------------------|-------------------------------------------------|-----------------------------------------------------|
| Detector: | heater, and autosample Quattro Micro Mass Sp | |
| Mode: | MRM | pectrometer |
| Source: | | |
| | Negative Electrospray | |
| LM 1 resolution: | 10.0 | |
| HM 1 resolution | 10.0 | |
| Ion energy 1: | 1.0 | |
| Entrance: | 5 | |
| Collision: | 2 | |
| Exit: | 5 | |
| LM 2 resolution: | 14 | |
| HM 2 resolution: | 14 | |
| Ion energy 2: | 2.0 | |
| Multiplier (V): | 650 | |
| Capillary (kV): | 2.0 | |
| Cone (V): | 15 | |
| Extractor (V): | 0 | |
| RF lens (V): | 0 | |
| Source temperature: | 130°C | |
| Desolvation temperature: | 350°C | |
| MS Method: | | |
| Mode: | MRM, 2 transitions | |
| Time: | 0-10.0 min | |
| Ch1: | $413.00 \rightarrow 369.00$ | |
| CIII. | 413.00 → 369.00 Dwell: | 0.25 sec |

Ammonium Perfluorooctanoate: Age Effect on the

Plasma Concentration in Post-Weaning Rats Following Oral Gavage

DuPont-13267

Ch2:

Collision energy:

15 eV

 $463.00 \rightarrow 419.00$

Dwell:

0.25 sec

Collision energy:

15 eV

4. Chromatographic Methods

Method:

Plasma concentration analysis

Column:

Waters Xterra MS C18, 2.1x30mm, 2.5 µm

Column temperature:

Ambient

Mobile phases:

A: 50 mM Ammonium Acetate

B: Acetonitrile

Gradient:

| Time (min) | %B |
|------------|-----|
| 0.0 | 10 |
| 0.5 | 10 |
| 10.0 | 100 |
| 10.9 | 100 |
| 11.0 | 10 |

Flow rate:

0.25 mL/min

Stop time:

11.0 min

Injection volume:

5 µL

F. Statistical Analysis

Group data is represented by mean, standard deviation, standard error, and percent coefficient of variation. Statistical significance was assessed by the Student's t-test. Rejection of data is based on Chauvenet's criterion. (7)

RESULTS AND DISCUSSION

A. **Dose Verification** (Appendix A)

The concentrations of the test substance in the dose solutions were verified by LC/MS method as described in Materials and Methods, Section E. Standards for dose verification and the dose solutions were prepared by different personnel. The determined mean PFOA concentrations of the dose solutions were all within 20% of the targeted concentration (2.5 mg/mL, Appendix A).

B. Plasma Concentration of PFOA (Table 1, Figures 1-3, Appendix B)

1. Male Rats

The plasma concentrations of PFOA in the male rats are shown in Table 1. Their relationship with the age of the male rats is plotted in Figure 1. The PFOA plasma concentration was $7.32 \pm 1.01~\mu g/mL$ (mean \pm SD) in 4-weeks old male rats. In contrast, a higher PFOA concentration (39.24 \pm 3.89 $\mu g/mL$) was observed in the plasma of 5-week old male rats. The mean PFOA plasma concentrations in 6-,7-, and 8-week old male rats were in a range of 37.12 to 43.19 $\mu g/mL$, which, based on the results of unpaired t-test at 99% confidence interval, were not significantly different from the plasma concentration of PFOA in the 5-week old male rats.

2. Female Rats

The plasma concentrations of PFOA in the female rats are shown in Table 1. Their relationship with the age of the female rats is plotted in Figure 2. The plasma concentration of PFOA in 4-week old female rats was $2.68 \pm 0.64 \,\mu g/mL$, which was significantly higher (p<0.01) than the PFOA concentrations (mean ranged from 0.57 to 1.18 $\,\mu g/mL$) in the elder females that were 5 to 8 weeks old.

3. Comparison between Male and Female Rats

PFOA plasma concentrations in male and female rats are shown in Figure 3. PFOA plasma concentrations were higher in the males than in the females for all age groups. The most dramatic sex-dependent difference in PFOA concentration was observed for rats that were 5 weeks and older. The ratios of PFOA concentrations between male and female rats that were 5 to 8 weeks old were all greater than 34, whereas the ratio was only 2.7 for the 4-week old rats.

CONCLUSIONS

Age has a significant effect on the plasma concentrations of PFOA in both male and female rats following oral dosing with 10 mg/kg PFOA. The sex-related difference in PFOA plasma concentration is most pronounced for rats that are older than 5 weeks, with male rats having PFOA plasma levels more than 34-fold higher than female rats.

RECORDS AND SAMPLE STORAGE

Specimens (if applicable), raw data, and the final report will be retained at Haskell Laboratory, Newark, Delaware, or at Iron Mountain Records Management, Wilmington, Delaware.

REFERENCES

- 1. Ohmori K., Kudo, H., Katayama, K., and Kawashima, Y. (2003). Comparison of the toxicokinetics between perfluorocarboxylic acids with different carbon chain length. Toxicology 184, 135-140.
- 2. Vanden Heuvel, J.P., Davis, J.W., Sommers, R., and Peterson, R.E. (1992) Renal excretion of perfluorooctanoic acid in male rats: inhibitory effect of testosterone. J. Biochem. Toxicol. 7, 31-36.
- 3. Kudo, N., Suzuki, E., Katakura, M., Ohmori, K., Noshiro, R., and Kawashima, Y. (2001) Comparison of the elimination between perfluorinated fatty acids with different carbon chain length in rats. Chem. Biol. Interact. 134, 203-216.
- 4. Ylinen, M., Hanhijarvi, H., Jaakonaho, J., and Peura, P. (1989) Stimulation by oestradiol of the urinary excretion of perfluorooctanoic acid in the male rat. Pharmacol. Toxicol. 65, 274-277.
- 5. DuPont Haskell Laboratory (2003). Perfluorooctanoic Acid: Toxicokinetics in the Rat. Unpublished report, DuPont-7473.
- 6. Vanden Heuvel, J.P., Kuslikis, B.I., Van Rafelghem, M.J., and Peterson, R.E. (1991). Tissue distribution, metabolism, and elimination of perfluorooctanoic acid in male and female rats. J. Biochem. Toxicol. 6, 83-92.
- 7. Taylor, J.R. (1982). In *An Introduction to Error Analysis*. E.D. Commins, Editor, Oxford University Press.



TABLES

TABLES

EXPLANATORY NOTES

ABBREVIATIONS:

SD

standard deviation

Plasma concentration in rats 24 hours post-dose following oral gavage with Table 1: 10 mg/kg PFOA.

| | Ago | Ms | iles | Fem | ales |
|---------------------------|-----------------------|------------------------------------------|--------------------------------------|---------------------------------------|---------------------------------------|
| Group | Age (weeks) | Mean | SD | Mean | SD |
| I II III IV V | 4 5 6 7 8 | 7.32 39.24 43.19 37.12 38.55 | 1.01 3.89 3.79 4.07 5.44 | 2.68 1.13 1.18 0.57 0.81* | 0.64 0.46 0.52 0.29 0.27* |

Data expressed as µg/mL.

^{*} According to Chauvenet's criterion, ⁽⁷⁾ the value of PFOA plasma concentration obtained from rat 511 (2.66 µg/mL) was not included for the calculation of the mean and the SD.

FIGURES

Figure 1: Plasma concentration in male rats 24 hours post-dose following oral gavage with 10 mg/kg PFOA.

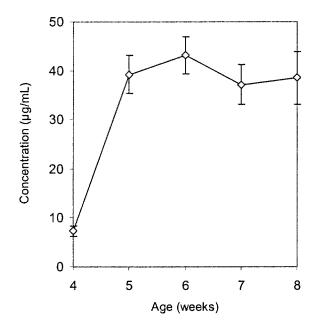


Figure 2: Plasma concentration in female rats 24 hours post-dose following oral gavage with 10 mg/kg PFOA.

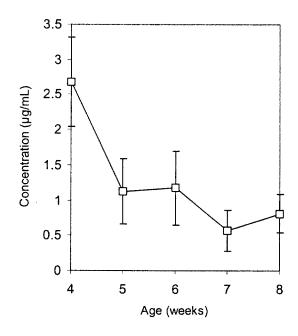
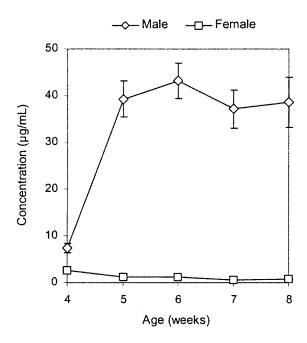


Figure 3: Plasma concentration in male and female rats 24 hours post-dose following oral gavage with 10 mg/kg PFOA.



APPENDICES

APPENDICES

EXPLANATORY NOTES

ABBREVIATIONS:

| LOQ | limit of quantitation |
|------|----------------------------------|
| SD | standard deviation |
| SE | standard error |
| % CV | percent coefficient of variation |

APPENDIX A

Dose Verification Data

| Number of Replicates | Description | mg/mL |
|----------------------|----------------------------------------------------|--------------|
| Dose Solution I – C | Froun I | |
| 1 | 250 ng/mL dilution – 3A | 2.64 |
| 2 | 250 ng/mL dilution – 3A | 3.01 |
| 3 | 250 ng/mL dilution – 3A | 2.70 |
| 4 | 250 ng/mL dilution – 3B | 2.83 |
| 5 | 250 ng/mL dilution – 3B | 2.60 |
| 6 | 250 ng/mL dilution – 3B | 2.84 |
| 7 | _ | |
| | 250 ng/mL dilution – 3C | 2.80 |
| 8 | 250 ng/mL dilution – 3C | 2.75 |
| 9 | 250 ng/mL dilution – 3C | 2.78 |
| Mean | | 2.77 |
| SD | | 0.12 |
| SE | | 0.04 |
| % CV | | 4.45 |
| Dose Solution II - | Group II | |
| 1 | 250 ng/mL dilution – 3A | 2.29 |
| 2 | 250 ng/mL dilution – 3A | 2.06 |
| 3 | 250 ng/mL dilution – 3A | 1.93 |
| 4 | 250 ng/mL dilution – 3B | 2.20 |
| 5 | _ | |
| | 250 ng/mL dilution – 3B | 2.22 |
| 6 | 250 ng/mL dilution – 3B | 1.87 |
| 7 | 250 ng/mL dilution – 3C | 1.77 |
| 8 | 250 ng/mL dilution – 3C | 1.86 |
| 9 | 250 ng/mL dilution – 3C | 1.85 |
| Mean | | 2.01 |
| SD | | 0.19 |
| SE | | 0.06 |
| % CV | | 9.47 |
| Dose Solution III - | Group III | |
| 1 | 250 ng/mL dilution – 3A | 2.69 |
| 2 | 250 ng/mL dilution – 3A | 2.65 |
| 3 | 250 ng/mL dilution – 3A | 2.62 |
| 4 | 250 ng/mL dilution – 3B | 2.59 |
| 5 | 250 ng/mL dilution – 3B | 2.82 |
| 6 | 250 ng/mL dilution – 3B | 2.55 |
| 7 | 250 ng/mL dilution – 3B | 2.53 |
| | | |
| 8 ' 9 | 250 ng/mL dilution – 3C 250 ng/mL dilution – 3C | 2.73 2.76 |
| | - | |
| Mean | | 2.67 |
| SD | | 0.09 |
| SE | | 0.03 |
| % CV | | 3.32 |

| Number of | | * * * * * * * * * * * * * * * * * * * * |
|---------------------|-------------------------|-----------------------------------------|
| Replicates | Description | mg/mL |
| | | |
| Dose Solution IV - | | |
| 1 | 250 ng/mL dilution - 3A | 2.49 |
| 2 | 250 ng/mL dilution - 3A | 2.47 |
| 3 | 250 ng/mL dilution – 3A | 2.32 |
| 4 | 250 ng/mL dilution - 3B | 2.51 |
| 5 | 250 ng/mL dilution 3B | 2.51 |
| 6 | 250 ng/mL dilution - 3B | 2.43 |
| 7 | 250 ng/mL dilution - 3C | 2.63 |
| 8 | 250 ng/mL dilution - 3C | 2.62 |
| 9 | 250 ng/mL dilution – 3C | 2.56 |
| Mean | | 2.50 |
| SD | | 0.09 |
| SE | | 0.03 |
| % CV | | 3.74 |
| Dose Solution V – C | Group V | |
| 1 | 250 ng/mL dilution – 3A | 2.28 |
| 2 | 250 ng/mL dilution – 3A | 2.19 |
| 3 | 250 ng/mL dilution – 3A | 2.11 |
| 4 | 250 ng/mL dilution – 3B | 2.18 |
| 5 | 250 ng/mL dilution - 3B | 1.78 |
| 6 | 250 ng/mL dilution - 3B | 2.07 |
| 7 | 250 ng/mL dilution – 3C | 1.85 |
| 8 | 250 ng/mL dilution – 3C | 2.20 |
| 9 | 250 ng/mL dilution – 3C | 2.21 |
| Mean | | 2.10 |
| SD | | 0.17 |
| SE | | 0.06 |
| % CV | | 8.13 |
| Dose Solutions I-V | | |
| Mean | | 2.41 |
| SD | | 0.34 |
| SE SE | | 0.11 |
| % CV | | 14.19 |
| / 0 C · | | 4 1147 |

APPENDIX B

Plasma Concentration Data

Males

| Animal Number | μg/mL |
|----------------------------------------------------|----------------------------------------------------------------------------------------|
| Group I | |
| 101 | 6.01 |
| 102 | 6.20 |
| 103 | 8.31 |
| 104 | 6.38 |
| 105 | 6.64 |
| 106 | 7.22 |
| 107 | 8.45 |
| 108 | 7.19 |
| 109 | 8.11 |
| Mean | 7.32 |
| SD | 1.01 |
| SE | 0.32 |
| % CV | 13.86 |
| Group II | |
| 201 | 33.40 |
| 202 | 39.43 |
| 203 | 46.42 |
| 204 | 41.89 |
| 205 | 38.72 |
| 206 | 37.35 |
| 207 | 42.64 |
| 208 | 35.11 |
| 209 | 40.96 |
| 210 | 36.45 |
| SE | 1.23 |
| % CV | 9.90 |
| Group III 301 302 303 304 305 306 307 308 309 310 | 38.14 39.95 42.00 45.27 41.38 43.46 48.23 50.44 40.99 42.01 |
| Mean | 43.19 |
| SD | 3.79 |
| SE | 1.20 |
| % CV | 8.77 |

Males (continued)

| Animal Number | μg/mL |
|---------------|-------|
| | |
| Group IV | |
| 401 | 30.49 |
| 402 | 31.28 |
| 403 | 40.22 |
| 404 | 41.41 |
| 405 | 36.76 |
| 406 | 35.14 |
| 407 | 38.70 |
| 408 | 35.96 |
| 409 | 38.36 |
| 410 | 42.93 |
| | 27.12 |
| Mean | 37.12 |
| SD | 4.07 |
| SE | 1.29 |
| % CV | 10.97 |
| Group V | |
| 501 | 32.66 |
| 502 | 37.93 |
| 503 | 46.78 |
| 504 | 27.47 |
| 505 | 38.05 |
| 506 | 39.94 |
| 507 | 41.65 |
| 508 | 38.03 |
| 509 | 39.16 |
| 510 | 43.85 |
| 310 | 12.00 |
| Mean | 38.55 |
| SD | 5.44 |
| SE | 1.72 |
| % CV | 14.10 |
| /U C + | |

Females

| Animal Number | μg/mL |
|---------------|--------------|
| C 1 | |
| Group I | 2.05 |
| 111 | 3.05 2.41 |
| 112 113 | 2.52 |
| 114 | 2.27 |
| 115 | 2.87 |
| 116 | 3.33 |
| 117 | 1.67 |
| 118 | 2.13 |
| 119 | 3.90 |
| 120 | 2.67 |
| 120 | 2.07 |
| Mean | 2.68 |
| SD | 0.64 |
| SE | 0.20 |
| % CV | 23.77 |
| | |
| Group II | |
| 211 | 0.98 |
| 212 | 1.18 |
| 213 | 0.81 |
| 214 | 1.79 |
| 215 | 1.65 |
| 216 | 0.76 |
| 217 | 1.47 |
| 218 | 0.69 |
| 219 | 0.43 |
| 220 | 1.52 |
| Mean | 1.13 |
| SD | 0.46 |
| SE SE | 0.15 |
| % CV | 41.13 |
| 70 C V | 11.13 |
| Group III | |
| 311 | 1.39 |
| 312 | 0.94 |
| 313 | 0.89 |
| 314 | 1.92 |
| 315 | 0.66 |
| 316 | 1.39 |
| 317 | 1.06 |
| 318 | 0.84 |
| 319 | 0.56 |
| 320 ` | 2.12 |
| | |
| Mean | 1.18 |
| SD | 0.52 |
| SE | 0.17 |
| % CV | 44.43 |
| | |

Females (continued)

| Animal Number | μg/mL |
|---------------|-------|
| 0 131 | |
| Group IV | 0.66 |
| 411 | 0.66 |
| 412 | 0.70 |
| 413 | 0.21 |
| 414 | 0.51 |
| 415 | 0.75 |
| 416 | 0.43 |
| 417 | 1.09 |
| 418 | LOQ |
| 419 | LOQ |
| 420 | 0.23 |
| Mean | 0.57 |
| SD | 0.29 |
| SE | 0.09 |
| % CV | 51.00 |
| Group V | |
| 511* | 2.66 |
| 512 | 0.87 |
| 513 | 0.63 |
| 514 | 0.95 |
| 515 | 1.36 |
| 516 | 0.67 |
| 517 | 0.64 |
| 518 | 0.89 |
| 519 | 0.50 |
| 520 | LOQ |
| Mean | 0.81 |
| SD | 0.27 |
| SE | 0.09 |
| % CV | 33.17 |

^{*} According to Chauvenet's criterion, (7) the value of PFOA plasma concentration obtained from rat 511 was not included for the calculation of the mean and the SD.